

essentially of residues 51 to 404 of PDK1 wherein Zaa represents a negatively charged amino acid residue for use in medicine.

32. (Twice Amended) A kit of parts for identifying a compound wherein the kit comprises PDK1 and a polypeptide comprising the amino acid sequence SEQ ID NO:25: Phe/Tyr-Xaa-Xaa-Phe/Tyr-Zaa-Phe/Tyr wherein Zaa represents a negatively charged amino acid residue.

33. (Amended) A method of making a preparation that comprises PDK1 and a polypeptide which comprises the amino acid sequence SEQ ID NO:25: Phe/Tyr-Xaa-Xaa-Phe/Tyr-Zaa-Phe/Tyr wherein substantially pure PDK1 is mixed with a substantially pure polypeptide which comprises the amino acid sequence SEQ ID NO:25: Phe/Tyr-Xaa-Xaa-Phe/Tyr-Zaa-Phe/Tyr.

After the claims, replace the previously filed Sequence Listing pages with replacement SEQUENCE LISTING pages 1-26 enclosed herewith.

REMARKS

Nucleotide Sequence Disclosure

In response to the notification of defective response, and in compliance with the sequence rules, applicants have enclosed the following:

1. A computer readable form (CRF) copy of a Sequence Listing in the form of a 3 1/2" diskette;
2. A paper copy of the replacement Sequence Listing, pages 1-26; and
3. A statement that the content of the paper and computer readable form are the same and include no new matter.

Each of the sequences presented in the Sequence Listing were present in the application as originally filed. Each of

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claims 1, 2, 4, 8, 10, 11, 14, 15, 22, 24, 30, 32 and 33 has been amended to include reference to a SEQ ID NO, where appropriate. Accordingly no new matter is involved. Applicants respectfully request that the Sequence Listing be entered and maintain that the application now complies with the sequence rules.

Respectfully submitted,

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Date

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Marked-Up Version of Claim(s)

1. (Amended) A method of altering the substrate specificity of phosphoinositide-dependent protein kinase 1 (PDK1) wherein the said PDK1 is exposed to an interacting polypeptide which comprises the amino acid sequence SEQ ID NO:25: Phe/Tyr-Xaa-Xaa-Phe/Tyr-Zaa-Phe/Tyr wherein Zaa represents a negatively charged amino acid residue.

2. (Amended) A preparation comprising PDK1 and an interacting polypeptide which comprises the amino acid sequence SEQ ID NO:25: Phe/Tyr-Xaa-Xaa-Phe/Tyr-Zaa-Phe/Tyr wherein Zaa represents a negatively charged amino acid residue characterised in that the said preparation is substantially free of polypeptides with which PDK1 is present or associated in a cell in which it is naturally found other than [a] said interacting polypeptide or a substrate for PDK1.

4. (Amended) A method of phosphorylating a residue corresponding to the [underlined] italicized residue in a substrate polypeptide with an amino acid sequence corresponding to the consensus sequence SEQ ID NO:30: Phe/Tyr-Xaa-Xaa-Phe/Tyr- [Ser/Thr] *Ser/Thr*-Phe/Tyr wherein (1) a preparation comprising PDK1 and a polypeptide which comprises the amino acid sequence SEQ ID NO:25: Phe/Tyr-Xaa-Xaa-Phe/Tyr-Zaa-Phe/Tyr or (2) PDK1 derivable by a method of altering the substrate specificity of phosphoinositide-dependent protein kinase 1 (PDK1) wherein the said PDK1 is exposed to a polypeptide which comprises the amino acid sequence SEQ ID NO:25: Phe/Tyr-Xaa-Xaa-Phe/Tyr-Zaa-Phe/Tyr is used wherein Zaa represents a negatively charged amino acid residue.

8. (Amended) A method of identifying a compound that modulates the activity of PDK1 wherein the said PDK1 is exposed to the said compound in the presence of a polypeptide comprising the amino acid sequence SEQ ID NO:25: Phe/Tyr-Xaa-

Xaa-Phe/Tyr-Zaa-Phe/Tyr wherein Zaa represents a negatively charged amino acid residue.

9. (Amended) A method according to claim 8 comprising the step of measuring the activity of the said PDK1 in the presence of more than one concentration of the compound wherein the said PDK1 is or has been exposed to a polypeptide which comprises the amino acid sequence SEQ ID NO:25: Phe/Tyr-Xaa-Xaa-Phe/Tyr-Zaa-Phe/Tyr.

10. (Twice Amended) A method according to claim 9 wherein said compound is capable of modulating the interaction between a polypeptide which comprises the amino acid sequence SEQ ID NO:25: Phe/Tyr-Xaa-Xaa-Phe/Tyr-Zaa-Phe/Tyr and PDK1.

11. (Amended) A method of identifying a compound that is capable of altering the substrate specificity of PDK1 wherein the ability of the said PDK1 to phosphorylate a residue corresponding to the [underlined] italicized residue in a polypeptide with an amino acid sequence corresponding to the consensus sequence SEQ ID NO:30: Phe/Tyr-Xaa-Xaa-Phe/Tyr-[Ser/Thr] Ser/Thr-Phe/Tyr is measured, and is increased in the presence of the said compound.

14. (Amended) A protein kinase derivable from mammalian brain wherein said protein kinase is capable of phosphorylating a residue corresponding to the [underlined] italicized residue in a polypeptide with an amino acid sequence corresponding to the consensus sequence SEQ ID NO:30: Phe/Tyr-Xaa-Xaa-Phe/Tyr-[Ser/Thr] Ser/Thr-Phe/Tyr, for example Ser473 of PKB α in the presence of PtdIns(3,4,5)P₃, wherein the said protein kinase is eluted from Heparin-Sepharose by at least 0.75M NaCl at pH 7.5 and is capable of binding to an antibody reactive with PDK1.

15. (Amended) A polypeptide which comprises the amino acid sequence SEQ ID NO:25: Phe/Tyr-Xaa-Xaa-Phe/Tyr-Zaa-Phe/Tyr wherein said polypeptide is not full-length PRK2, PRK1

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or PKC ζ and wherein Zaa is a negatively charged amino acid that is not phosphoserine or phosphothreonine.

22. (Amended) A cell containing a recombinant nucleic acid suitable for expressing PDK1 and a recombinant nucleic acid suitable for expressing a polypeptide comprising the amino acid sequence SEQ ID NO:25: Phe/Tyr-Xaa-Xaa-Phe/Tyr-Zaa-Phe/Tyr.

24. (Amended) A method of making a preparation comprising PDK1 and an interacting polypeptide comprising the amino acid sequence SEQ ID NO:25: Phe/Tyr-Xaa-Xaa-Phe/Tyr-Zaa-Phe/Tyr wherein PDK1 and the said interacting polypeptide are co-expressed in a cell as defined in claim 22.

30. (Amended) A polypeptide which comprises the amino acid sequence SEQ ID NO:25: Phe/Tyr-Xaa-Xaa-Phe/Tyr-Zaa-Phe/Tyr or a polypeptide consisting essentially of residues 51 to 404 of PDK1 or a fusion of a polypeptide consisting essentially of residues 51 to 404 of PDK1 wherein Zaa represents a negatively charged amino acid residue for use in medicine.

32. (Twice Amended) A kit of parts for identifying a compound wherein the kit comprises PDK1 and a [polypeptide] polypeptide comprising the amino acid sequence SEQ ID NO:25: Phe/Tyr-Xaa-Xaa-Phe/Tyr-Zaa-Phe/Tyr wherein Zaa represents a negatively charged amino acid residue.

33. (Amended) A method of making a preparation that comprises PDK1 and a polypeptide which comprises the amino acid sequence SEQ ID NO:25: Phe/Tyr-Xaa-Xaa-Phe/Tyr-Zaa-Phe/Tyr wherein substantially pure PDK1 is mixed with a substantially pure polypeptide which comprises the amino acid sequence SEQ ID NO:25: Phe/Tyr-Xaa-Xaa-Phe/Tyr-Zaa-Phe/Tyr.